

# FORAGE & GRAZING LANDS

## Seed Germination and Dormancy in Eastern Gamagrass

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### ABSTRACT

Eastern gamagrass [*Tripsacum dactyloides* (L.) L.] is an important native rangeland species and could be a more widely used perennial pasture grass. Stand establishment, however, is slow, and it can take 3 yr before a full stand is attained. Our objectives were to determine the caryopsis weight distribution, the relationship between caryopsis weight and percentage germination, the role of the cupulate fruit case in seed dormancy, and the force required to open the cupulate fruit case. Caryopses of 'Iuka' and 'Pete' eastern gamagrass were extracted from the cupulate fruit case by hand, individually weighed, separated into five weight classes, and germinated in a germination chamber. Differences in percentage observed germination and total potential germination at 7 and 14 d were attributed to caryopsis weight ( $P > 0.05$ ). No caryopsis weight  $\times$  cultivar interaction occurred, suggesting homogeneity of slopes for cultivars ( $P > 0.25$ ). An artificial fruit case (band) was fashioned from an extruded plastic tube and a caryopsis placed inside to examine the role of the fruit case in germination. Germination of unbanded caryopses (44.8%) was significantly greater than banded caryopses (6.3%,  $P \leq 0.01$ ). A lever and fulcrum were used to find the force required to open the fruit case. We concluded that seed dormancy was likely mechanical for these cultivars. Under natural conditions, the integrity of the fruit case must be reduced before germination will proceed normally. Light, alternating temperatures, freezing and thawing, fire, rodents, and soil microorganisms, to name a few, may be involved in the decay of the fruit case.

EASTERN GAMAGRASS is a perennial, warm-season bunchgrass occurring in various habitats across the eastern and central USA, Mexico, Central America, northern South America, and the Caribbean Islands (Newell and de Wet, 1974; de Wet et al., 1982). Typically, the inflorescence of eastern gamagrass consists of one or more spike-like racemes with staminate spikelets above and pistillate spikelets below (andromonoecism). Pistillate spikelets, borne on the basal one-fourth to one-third of the raceme, are usually solitary at each rachis node and contain one fertile floret. The indurate first glume and hard bony rachis internode form a cupule or fruit case that surrounds the pistillate spikelet. The second glume folds tightly inward and secures the pistillate spikelet, and at maturity, the caryopsis. Staminate spikelets, borne on the apical two-thirds to three-fourths of the raceme, occur in pairs at each rachis node and contain two staminate florets.

This grass has been recognized as one of the most

productive and palatable warm-season grasses of the southeastern USA (Dewald and Louthan, 1979). Fine et al. (1990) reported forage dry-matter production of 5.0 Mg ha<sup>-1</sup> in the first year of establishment and 22.6 Mg ha<sup>-1</sup> in the third year. They also reported that in vitro dry-matter digestibility at first harvest averaged 63.1%. Limited information is available on animal production with eastern gamagrass; however, Burns et al. (1991) reported average daily gains of 0.82 kg d<sup>-1</sup> under variable stocking rates, and Aiken and Springer (1994) reported average daily gains of 0.80 kg d<sup>-1</sup> using 3.0 head ha<sup>-1</sup>, 0.89 kg d<sup>-1</sup> using 4.9 head ha<sup>-1</sup>, and 0.75 kg d<sup>-1</sup> using 7.4 head ha<sup>-1</sup>.

Problems associated with stand establishment continue to limit the widespread use of eastern gamagrass. An early concern related to seed quality was determining the pure seed unit, which is impossible to do visually. The pure seed unit is a caryopsis enclosed by the cupulate fruit case. Removal of empty seed units, for example, sterile cupulate fruit cases, will improve seed quality. Ahring and Frank (1968) successfully removed empty seed units with air and floatation separation. The continuing concern related to seed quality is germination of the pure seed unit (intact fertile cupules). To germinate, the caryopsis within the cupule must imbibe water, and the embryo must grow and emerge from the cupule. Several chemical agents (potassium nitrate, sodium hypochlorite, gibberellic acid, and carbon dioxide) have been used unsuccessfully to stimulate seed germination (Ahring and Frank, 1968; Anderson, 1985). Cold-moist stratification apparently weakens the cupule enough to allow the caryopsis to germinate. Ahring and Frank (1968) reported that stratification at 5 to 10°C for 6 to 8 wk enhanced seed germination to 60 to 80% of all viable seeds. Anderson (1985) reported similar findings, except that germination percentages were lower for his seed lots. Ahring and Frank (1968) also found differences among seed lots for germination and response to prechill duration.

In contrast, naked caryopses (i.e., with the cupulate fruit case removed) germinate readily (Anderson, 1985). An alternative to planting intact cupules is to plant caryopses. An advantage of planting caryopses is that pastures could be seeded to known plant densities. At present time, however, the specialized equipment needed to hull gamagrass without damaging the caryopsis has not been developed.

Improving germination and alleviating seed dormancy of intact cupules could substantially enhance stand es-

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Abbreviations: RMSE, root mean square error.

tablishment. Thus, we need to understand the role the cupulate fruit case plays in dormancy of eastern gamagrass. The objectives of our study were to determine: (i) the caryopsis weight distribution, (ii) the relationship between caryopsis weight and percentage germination, (iii) the role of the cupulate fruit case in seed dormancy, and (iv) the force required to open the cupulate fruit case.

## MATERIALS AND METHODS

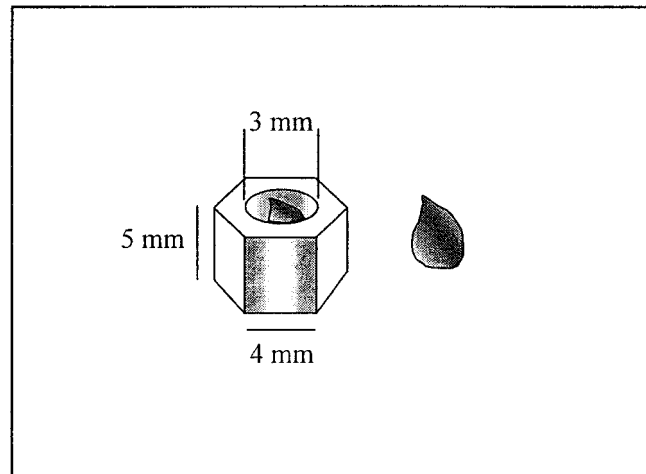
The cultivars of eastern gamagrass used in this study were Iuka, obtained from Mott Ranch, Inc., Pratt, KS, and Pete, obtained from Shepherd Farms, Inc., Clifton Hill, MO. The seed of each cultivar were stored  $\approx 3$  mo at ambient room temperatures prior to study.

### Caryopsis Weight Distribution

Seed of each cultivar were randomly separated into four samples, each containing 1000- seed units. Caryopses were extracted from each sample by hand and individually weighted and placed into five weight classes (1.0–10.0, >10.0–20.0, >20.0–30.0, >30.0–40.0, and >40.0 mg caryopsis<sup>-1</sup>). Caryopses within each class were weighed and counted. Mean caryopsis weight was calculated by dividing the total caryopses weight (mg) of each class by the number of caryopses in each class. Caryopsis weight distribution percentages (based on caryopses numbers) were calculated by dividing the number of caryopses in each class by the total number of caryopses in the sample. Dependent variables were analyzed using a completely random design. The cultivar Iuka was replicated three times, and Pete four times.

### Caryopsis Germination and Seedling Development

Fifty caryopses of each weight class were placed in  $7.0 \times 7.0 \times 2.5$ -cm plastic germination boxes on two layers of towel substrate moistened with 7 mL of distilled water. Germination boxes were placed in a seed germinator (Stultz Scientific Engineering Corp., Springfield, IL)<sup>1</sup> set for alternating temperatures of 20°C (dark) and 30 °C (light: fluorescent; photosynthetic photon flux density =  $9.0 \mu\text{mol m}^{-2} \text{s}^{-1}$ ) at 16 and 8 h, respectively. Cumulative germination counts were made at 7 and 14 d of "normal germinated seedlings" (Colbry et al., 1961). At the end of 14 d, the number of firm (nongerminated, apparently dormant) caryopses was recorded. Total potential germination was determined by adding the number of seeds observed that germinated and the number of remaining firm caryopses after 14 d. Data were converted to percentages before analysis. Germination percentage were not transformed prior to analysis, because residuals were distributed normally. Seedling shoot and root lengths were used as a measure of seedling vigor. Five random seedlings were measured for each weight class at the 7-d germination count. The experimental design was a randomized block replicated four times and repeated twice. The caryopsis weight  $\times$  cultivar interaction was used to test the heterogeneity of slopes between cultivars (Littell et al., 1991). The relationship between 14-d observed germination, total potential germination, and shoot and root



**Fig. 1.** Illustration of an artificial fruit case (band) fashioned from an extruded plastic hexagonal tube (barrel of an ink pen) and a naked caryopsis used to test for mechanical seed dormancy in eastern gamagrass. The picture is not drawn to scale. The dimensions of the band were 7 mm wide from face to face and 5 mm tall. Each of the six faces were 4 mm wide and 5 mm tall. The circular opening of the band was 3 mm in diameter and open to the top and bottom.

lengths and caryopsis weight were analyzed using regression analyses.

### Effect of Cupule Removal or Presence on Germination

We conducted an experiment to find the role of the cupulate fruit case on seed dormancy. An artificial fruit case (band) was fashioned from an extruded plastic hexagonal tube (barrel of an ink pen). The dimensions of the band were 7 mm wide from face to face and 5 mm tall. Each of the six faces were 4 mm wide and 5 mm tall. The circular opening of the band was 3 mm in diameter and open to the top and bottom. A caryopsis from the >30.0- to 40.0-mg weight class was placed inside the band (a caryopsis of this weight class fits snugly, without damage to the caryopsis, into the band), where the bottom of the caryopsis was flush with the bottom edge of the band. This allowed water to come into contact with the caryopsis when placed onto the paper towel substrate in the germination boxes. A banded caryopsis was paired with a naked caryopsis (Fig. 1) and placed in a germination box on two layers of towel substrate moistened with 7 mL of distilled water. The germination environment was the same as in the previous germination experiment. Germination percentages among cupule classes were analyzed using a paired *T*-test with 48 pairs. The experiment was repeated twice.

### Force Required to Open the Cupulate Fruit Case

An experiment was conducted to determine the effect of moist prechill on the force required to open the cupulate fruit case. A 91.4-cm lever and a fulcrum placed 30.5 cm from the cupulate fruit case was used to determine the force required to open the fruit case. A dissecting needle, attached to the short arm of the lever, was inserted 2 to 3 mm under the outer glume of the cupulate fruit case; at the other end of the lever hung a small bucket. Salt was added to the bucket until the fruit case hinge opened. The salt-filled bucket was weighted to the nearest kilogram and the weight multiplied by 2 (because the bucket hung on the long arm of the lever, Smith and Cooper, 1957). This product was multiplied by 9.807 N kg<sup>-1</sup> (Smith and Cooper, 1957) to find the force required to

<sup>1</sup> Mention of a trademark or a proprietary product does not constitute a guarantee or warranty of the product by USDA and does not imply approval to the exclusion of other suitable products. All programs and services of the USDA are offered on a nondiscriminatory basis without regard to race, color, national origin, religion, sex, age, marital status, or handicap.

open the cupulate fruit case. The relationship between duration of chilling and force required to open the cupulate fruit case was analyzed using regression analysis. The experiment consisted of four replications of five seeds each at 0, 1, 2, 4, and 8 wk of prechill at 0 to 5°C. The experiment was repeated twice.

## RESULTS AND DISCUSSION

### Caryopsis Weight Distribution

Caryopses of both cultivars displayed unimodal weight distributions (Table 1), and the caryopsis made up  $\approx 32\%$  of the weight of the intact fruit case. The mean caryopsis weight of Iuka was  $21.3 \pm 0.59$  mg (mean  $\pm$  SD), compared with  $23.3 \pm 1.55$  mg for Pete. Eastern gamagrass matures seed continuously from mid-June through mid-August, and the environmental conditions before, during, and after flowering may significantly influence variation in caryopsis weight (Roach and Wulff, 1987). Similarly, interrupting the reproduction cycle (as by seed harvest) during caryopsis filling may also increase the variation in caryopses weight as well as increase seed-borne diseases.

### Germination of Caryopses and Seedling Development

Differences among weight classes in observed germination percentages and total potential germination percentages at 7- and 14-d were attributed only to caryopsis weight. No caryopsis weight  $\times$  cultivar interaction ( $P > 0.25$ ) occurred, suggesting homogeneity of slopes for cultivars. Percentage 7- and 14-d observed germination and total potential germination of weight classed caryopses increased as caryopsis weight increased. The relationship between observed germination at 7-d ( $Y$ , %) and caryopsis weight ( $x$ , mg) was  $Y = 51.48 - 344.99 x^{-1}$ ,  $P < 0.01$ , RMSE = 6.44,  $R^2 = 0.84$ ; observed germination at 14 d ( $Y$ , %) and caryopsis weight ( $x$ , mg) was  $Y = 52.33 - 346.79 x^{-1}$ ,  $P < 0.01$ , RMSE = 6.64,  $R^2 = 0.83$  (Fig. 2A); and potential germination at 14 d ( $Y$ , %) and caryopsis weight ( $x$ , mg) was  $Y = 60.28 - 403.79 x^{-1}$ ,  $P < 0.01$ , RMSE = 7.00,  $R^2 = 0.86$  (Fig. 2B).

Differences in percentage firm (dormant) seed were attributed to caryopsis weight. The relationship between firm seed remaining at 14 d ( $Y$ , cm) and caryopsis weight

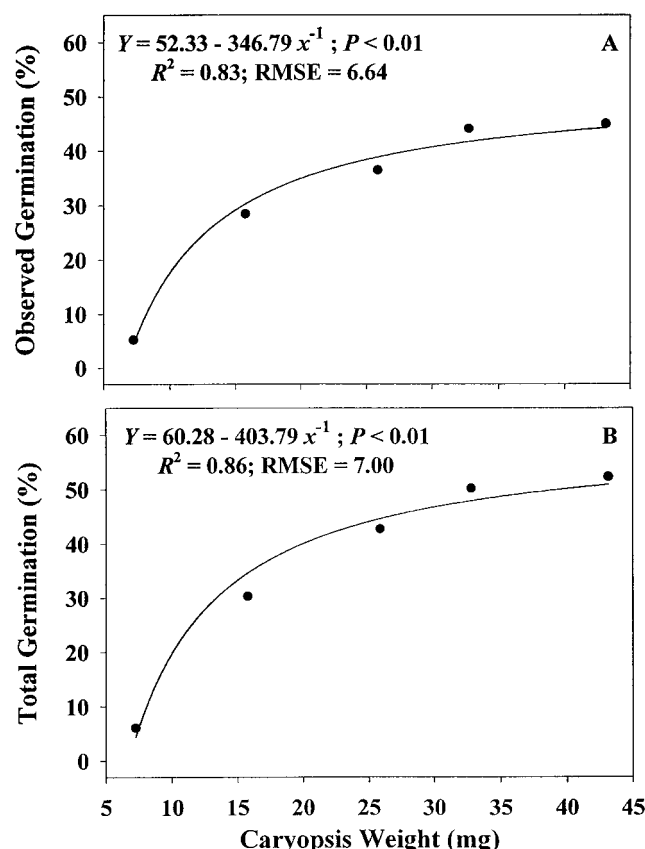


Fig. 2. Relationships of eastern gamagrass caryopsis weight and observed germination at the end of a 14-d germination period (A), and caryopsis weight and total potential germination (14 d observed germination plus firm seed remaining) (B). Each data point is the mean of eight experimental units.

( $x$ , mg) was  $Y = 7.16 - 51.62 x^{-1}$ ,  $P < 0.01$ , RMSE = 3.06,  $R^2 = 0.31$ .

Significant variation in root and shoot length was also attributed to caryopsis weight. Similarly, no caryopsis weight  $\times$  cultivar interaction occurred. The relationship between root length ( $Y$ , cm) and caryopsis weight ( $x$ , mg) was  $Y = -1.89 + 1.39 \ln x$ ,  $P < 0.01$ , RMSE = 0.51,  $R^2 = 0.75$  (Fig. 3A); and shoot length ( $Y$ , cm) and caryopsis weight ( $x$ , mg) was  $Y = -0.08 + 0.47 \ln x$ ,  $P < 0.01$ , RMSE = 0.24,  $R^2 = 0.61$  (Fig. 3B).

Significant variation in the percentages of caryopses with mold was attributed to cultivar, caryopsis weight, and caryopsis weight  $\times$  cultivar interactions. Thus, separate equations were estimated for each cultivar. The relationship between seeds with mold ( $Y$ , %) and caryopsis weight ( $x$ , mg) for Iuka was  $Y = 102.72 - 0.00011 x^{3.42}$ ,  $P < 0.02$ , RMSE = 12.90,  $R^2 = 0.54$  and for Pete was  $Y = 99.19 - 0.000019 x^{3.99}$ ,  $P < 0.01$ , RMSE = 15.68,  $R^2 = 0.74$ .

The observed and potential percentage germination of caryopses of eastern gamagrass are significantly influenced by caryopsis weight. Similar findings have been reported for other native North American grass species (Kneebone and Cremer, 1955; Springer, 1991). Environmental conditions reducing plant stress during caryopsis filling may lead to an increase in photosynthates that enter the caryopsis, resulting in a larger embryo and/or

Table 1. Caryopsis weight and distribution percentages (based on caryopses numbers) of *Tripsacum dactyloides* (cultivars 'Iuka' and 'Pete') by seed weight class.

		Distribution	
Weight class	Weight $\pm$ SD	Iuka	Pete
mg		%	
1.0–10.0	7.12 $\pm$ 0.84	13.2a†	9.0b
>10.0–20.0	15.78 $\pm$ 0.53	32.0a	26.5a
>20.0–30.0	26.05 $\pm$ 3.03	33.3a	41.0a
>30.0–40.0	32.94 $\pm$ 3.94	19.5a	18.6a
>40.0	43.13 $\pm$ 0.51	2.0a	4.9a
Non-classed‡	22.17 $\pm$ 1.43		

† Cultivar means within each seed weight class (row) followed by the same letter are not significantly different at  $P \leq 0.05$  [F-test].

‡ Average weight of non-classed caryopses of  $>1.0$  mg.

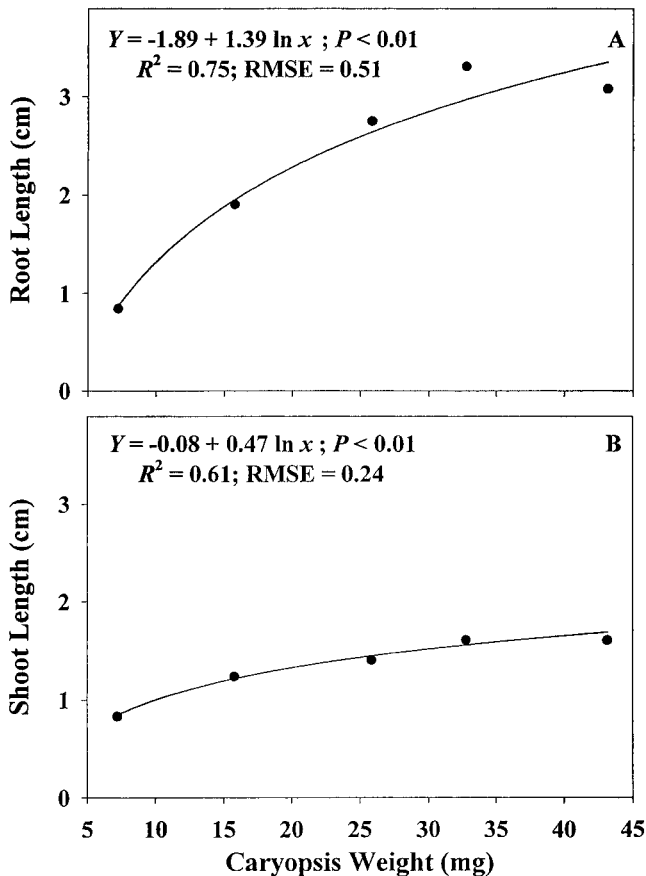


Fig. 3. Relationships of eastern gamagrass caryopsis weight and seedling root length (A) and caryopsis weight and seedling shoot length (B). Seedling measurements were made at end of the 7-d germination period. Each data point is the mean of 8 experimental units.

carbohydrate reserve. Larger caryopses tend to have more rapid and greater percentage germination and are less susceptible to attack from microorganisms than are smaller caryopses. Large caryopses also produce larger, more vigorous seedlings which are more likely to compete successfully for moisture and light than do seedlings from small seeds. The major cause of seedling death among prairie species is moisture stress (Blake, 1935).

#### Effect of Cupule Removal or Presence on Germination

The 7-d germination of caryopses banded with an artificial fruit case was significantly lower ( $P < 0.01$ ) than the unbanded control. Unbanded caryopses of the cultivar Iuka averaged  $37.5 \pm 7.1\%$  (mean  $\pm$  SE) compared with  $8.3 \pm 4.0\%$  for the banded caryopses. Similarly, unbanded caryopses of Pete averaged  $50.0 \pm 7.3\%$ , compared with  $4.2 \pm 2.9\%$  for the banded caryopses. For both cultivars, the germination percentages of the unbanded caryopses were similar to those found for the >30.0- to 40.0-mg weight seed class in the first germination experiment.

Seed dormancy in these cultivars of eastern gamagrass is likely mechanical in nature. Seed dormancy for the majority of species, where seed is defined in a broad sense, is associated with the structures surrounding the

embryo. The seed coat of grasses often times includes the glumes, lemma, and palea (Bewley and Black, 1982). Seed coat-imposed dormancy may interfere with water uptake or gaseous exchange; the coat may contain chemical inhibitors or act as a barrier against the escape of inhibitors from the embryo; it may modify the light that reaches the embryo; or it may exert a mechanical restraint (Bewley and Black, 1982). In some species, one or more of these actions may take place. Several grass genera have been reported with coat-imposed dormancy, however, the dormancy was associated with chemical that inhibit germination rather than mechanical restraint (Bewley and Black, 1982).

#### Force Required to Open the Cupulate Fruit Case

Fruit case integrity is, in part, a function of moisture availability, chilling temperatures, and time. The relationship of the force required to open a fruit case ( $Y$ , N) and the duration of prechill ( $x$ , wk) was  $Y = 4.05 + 2.54 e^{-x}$ ,  $P < 0.01$ , RMSE = 1.16,  $R^2 = 0.40$  (Fig. 4). Determined on the basis of the equation and data, the rate of decline in the force required to open the cupulate fruit case does not change significantly after 4 wk of prechill. Thus, a stratification period of at least 4 wk should be a sufficient pregermination treatment for these cultivars.

Under natural conditions, a caryopsis must first overcome the integrity of the cupulate fruit case to germinate. Given that the force required to open the fruit case does not change significantly after 4 wk of moist prechill, other factors such as light, alternating temperatures, freezing and thawing, fire, rodents, and soil microorganisms may be involved in the slow decay of the fruit case. If the caryopsis remains viable over a long enough period of time, and sufficient weathering occurs to the fruit case, germination should proceed as normal. However, given a long decay process, chances are likely that the enclosed caryopsis will succumb to microorganisms. Thus, the previously reported benefits of prechill treatment are not due primarily to immediate effects on the cupule integrity, but may likely result from a

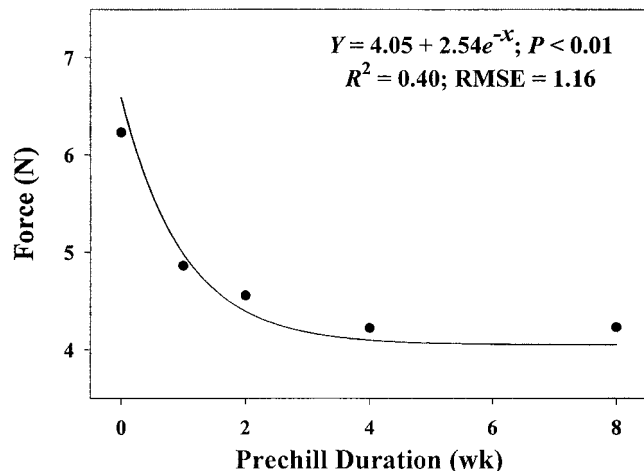


Fig. 4. Relationship of duration of moist prechill at 0 to 5 °C on the force required to open the cupulate fruit case of eastern gamagrass. Each data point is the mean of 10 experimental units.



predisposition to the cupule to more rapid deterioration from weathering processes.

## CONCLUSIONS

The germination of Pete and Iuka eastern gamagrass seed was affected by caryopsis weight, in that larger caryopses germinated better than smaller caryopses. These results are consistent with previously published results for several plant species. Seed dormancy of these two cultivars was attributed to the indurate first glume and hard bony rachis internode, which form the cupulate fruit case. The integrity of the tissue where the first glume and the rachis internode meet must be reduced before germination will proceed normally. Our research suggested that a minimum of 4-wk moist prechill at 0 to 5°C is required to overcome the integrity of the cupulate fruit case, and previous research has shown that 6 to 8 wk of moist prechill at 5 to 10°C is effective for increasing the germination of intact eastern gamagrass spikelets. Furthermore, winter plantings of unhulled eastern gamagrass in the southern Piedmont have resulted in initial stands that were equal to or greater to stands established in the spring with unhulled stratified seeds (Mueller et al., 2000). Thus, planting unhulled eastern gamagrass seed in winter should significantly improve stand establishment, provided that soil temperatures and moisture conditions are favorable for seed stratification. An alternative to winter seeding eastern gamagrass is to plant stratified seed in early spring if soil temperatures and moisture are favorable for establishment.

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